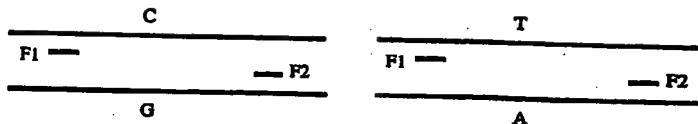
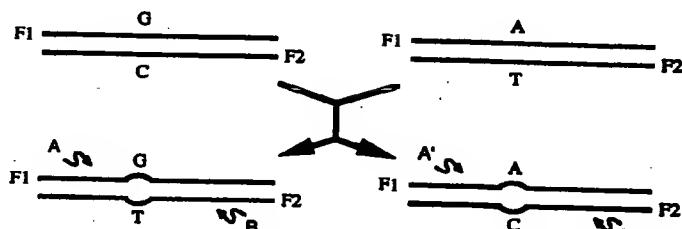


**Mismatch scanning Assay.
(Endo V / DNA Ligase)**

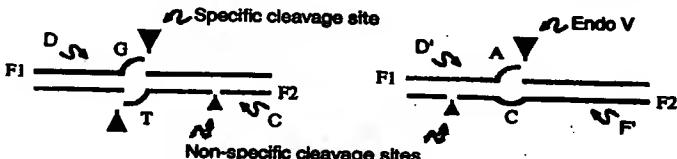
1. PCR amplify gene using primers with different fluorescent labels and *Taq* DNA polymerase.



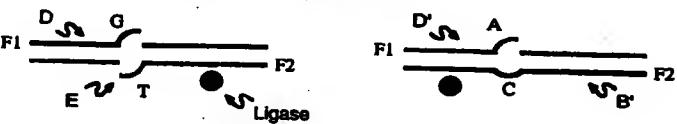
2. Denature and reanneal PCR products to form heteroduplexed DNA. (Homoduplexed products not shown).



3. Preferentially nick DNA one base to the 3' side of mismatches using thermostable Endonuclease V.



4. Add thermostable ligase to re-seal background nicks at perfect match regions.



5. Separate fluorescent products on a DNA sequencer (using length standards) to approximate site of mismatch.

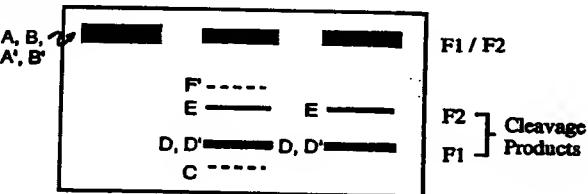


Figure 1

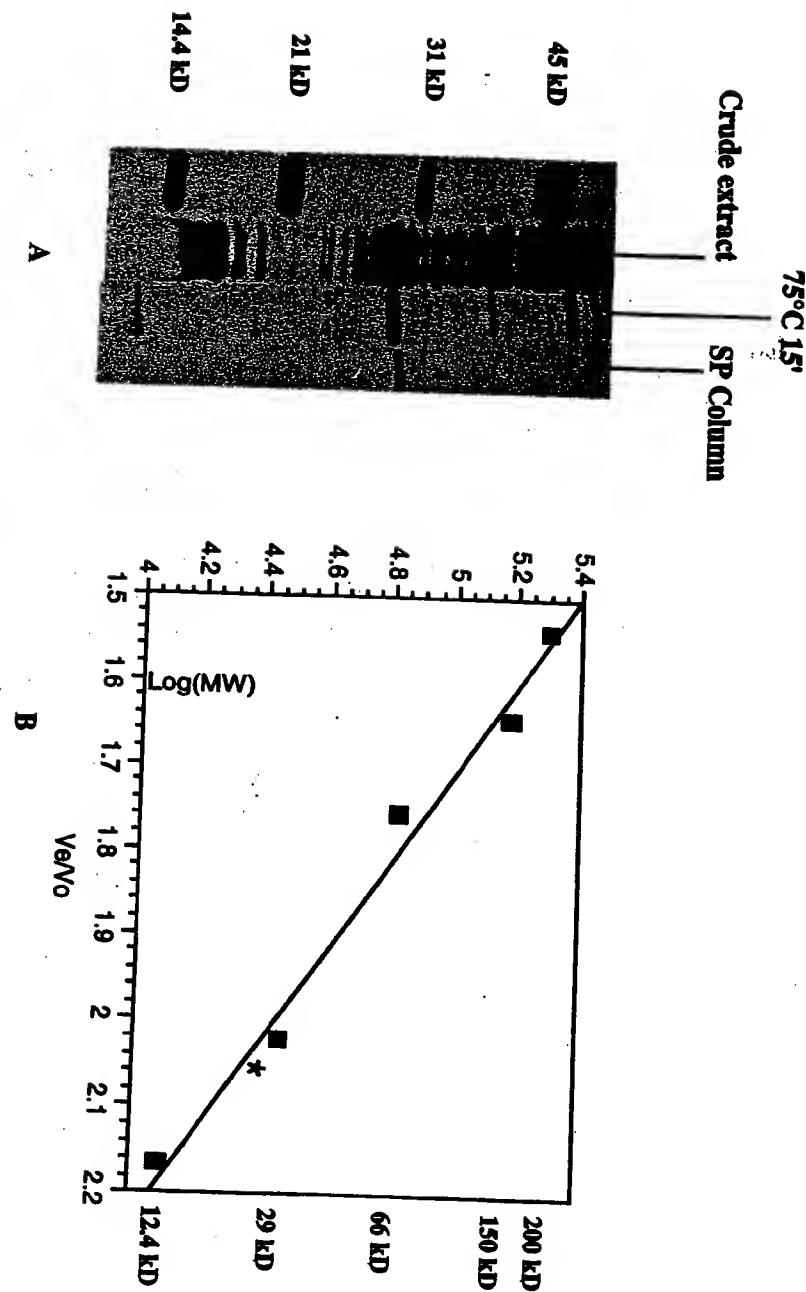


Figure 2

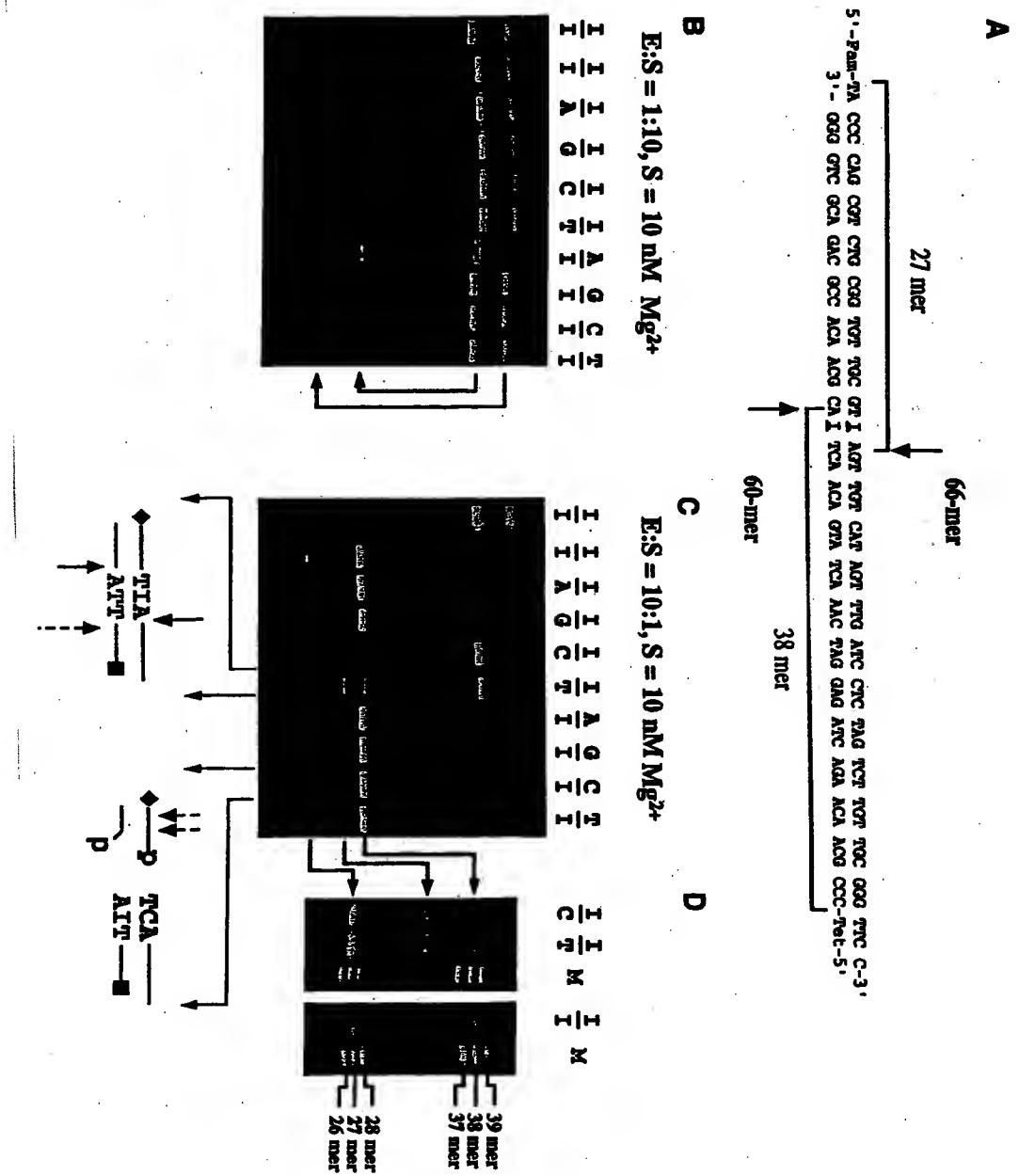
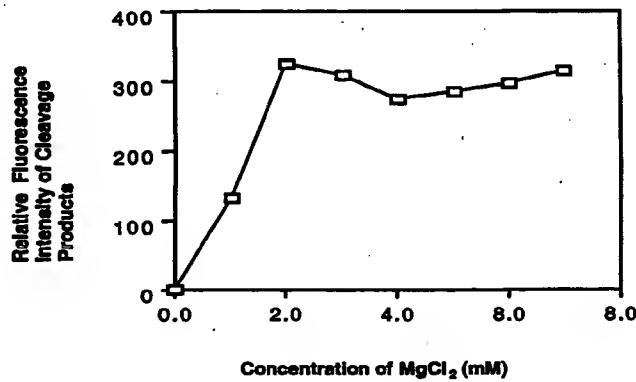


Figure 3

A



B

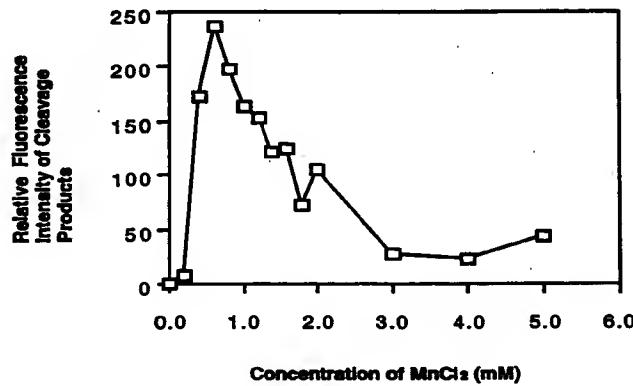


Figure 4

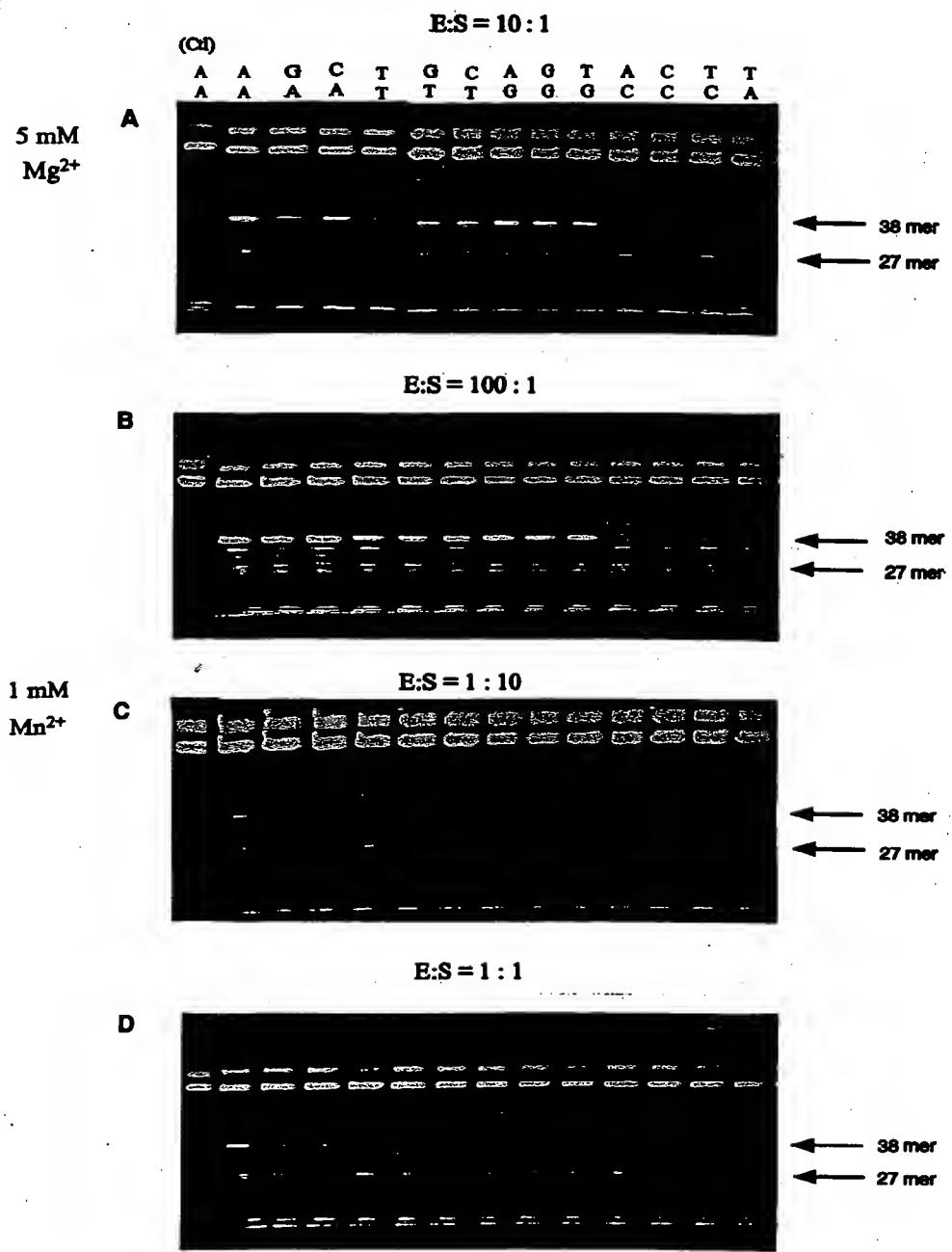


Figure 5

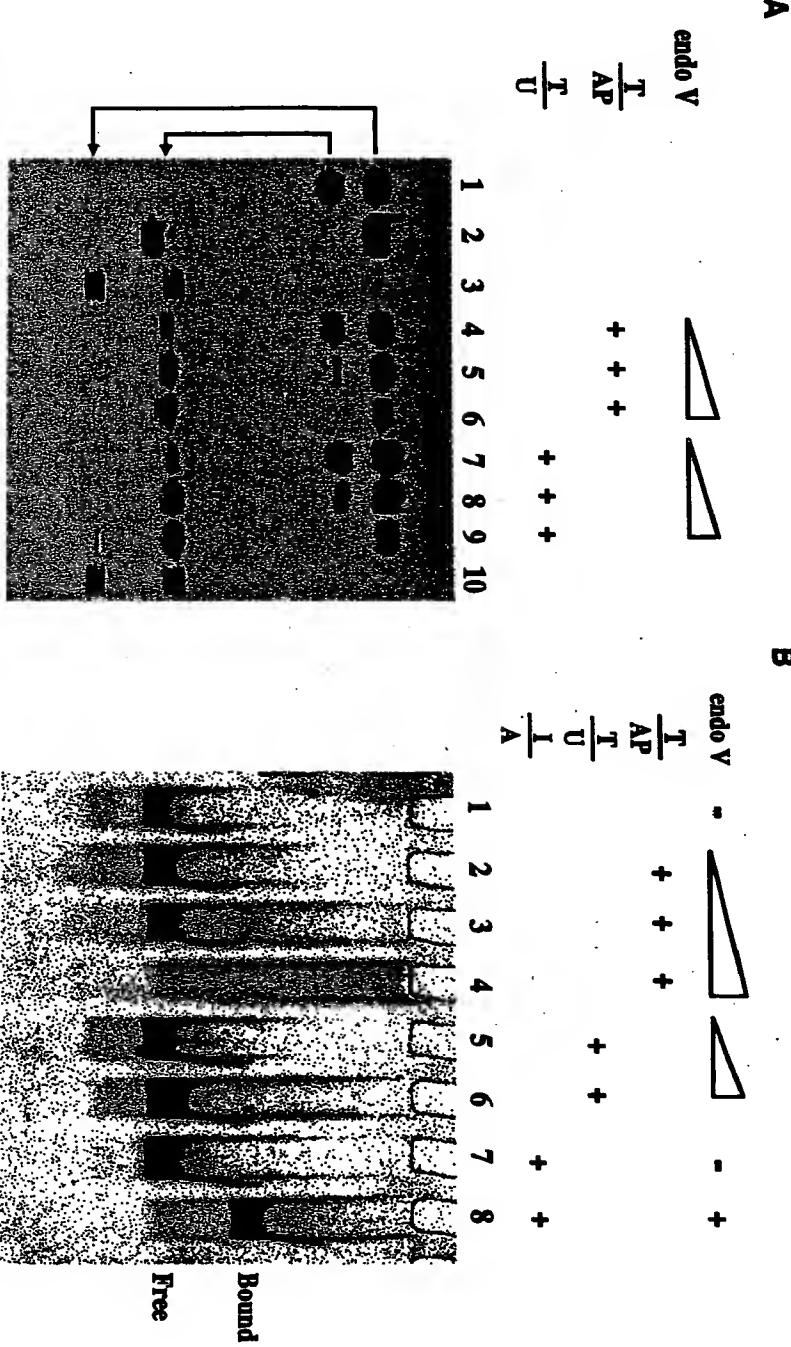


Figure 6

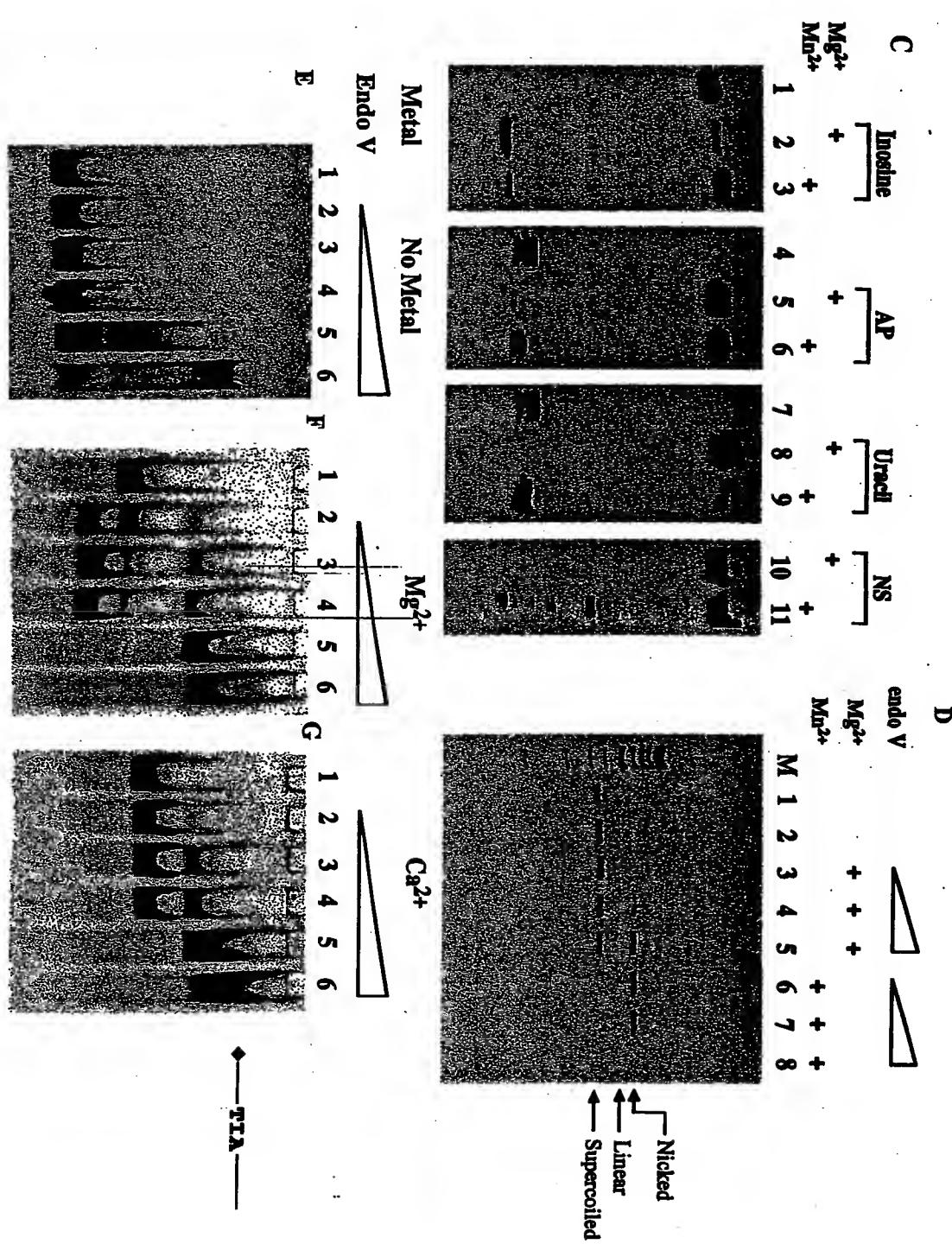


Figure 6 (cont.)

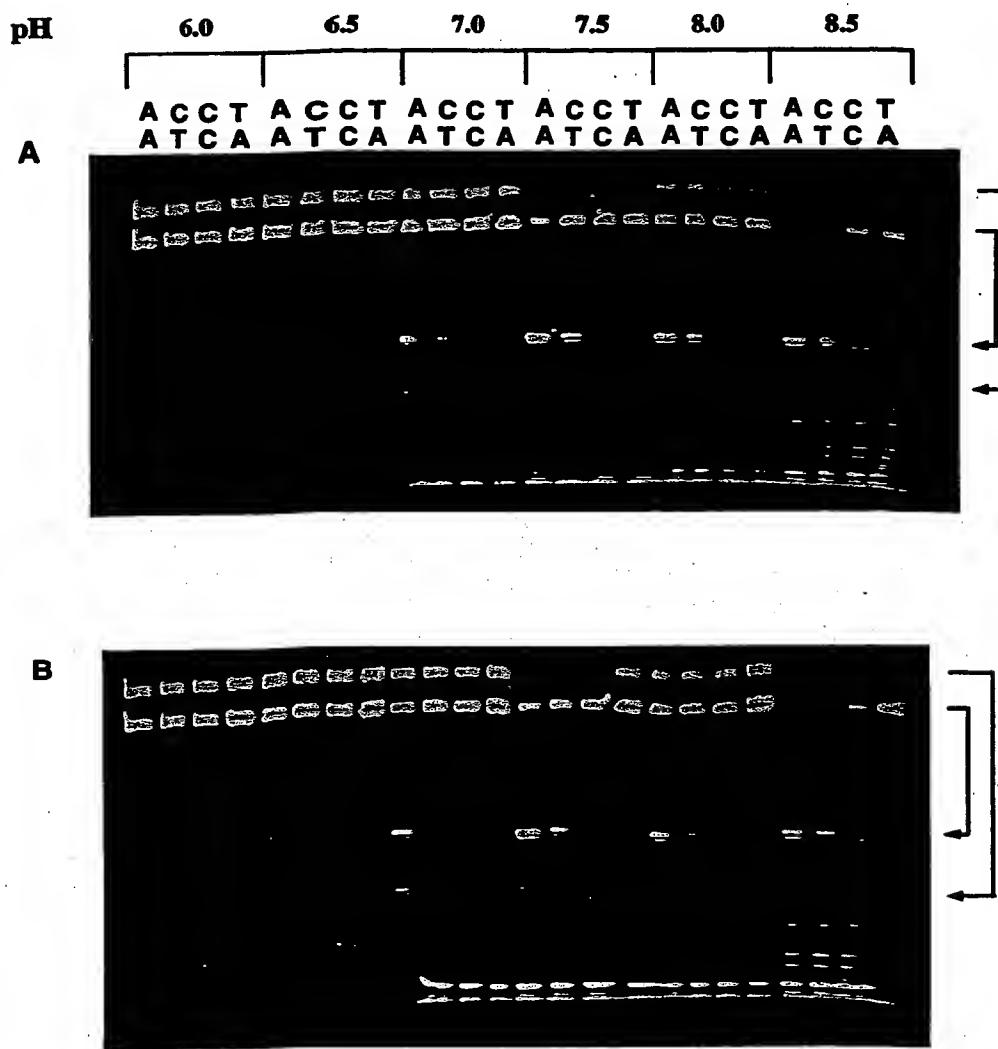


Figure 7

Effect of salt

NaCl/mM

0 50 100 150 200

Mg²⁺

Mn²⁺

Figure 8

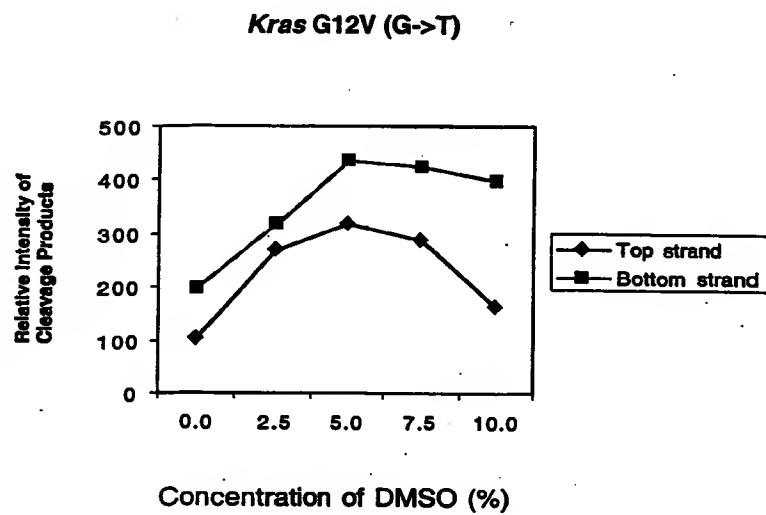
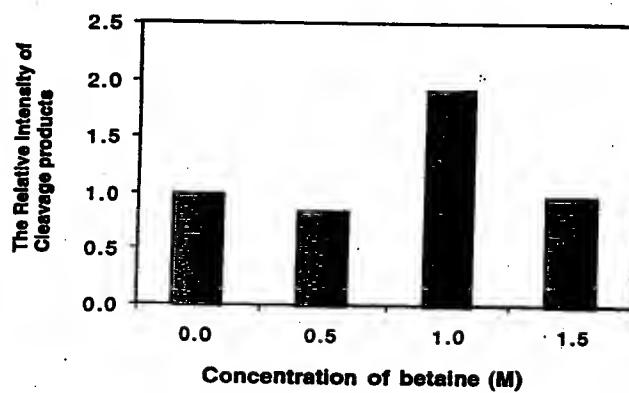


Figure 9

A

APC I1307K(T->A)



B

Kras G12V (G->T)

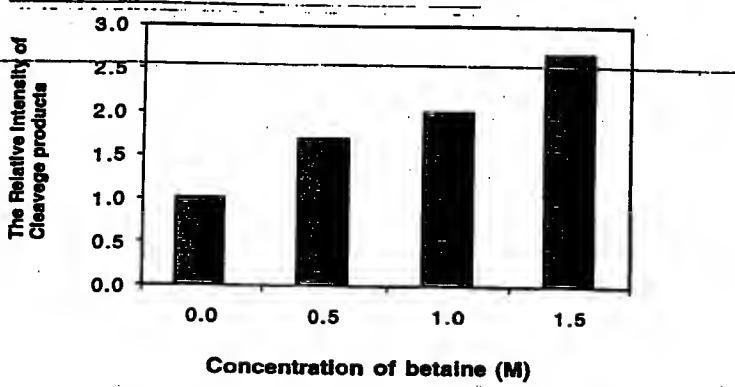
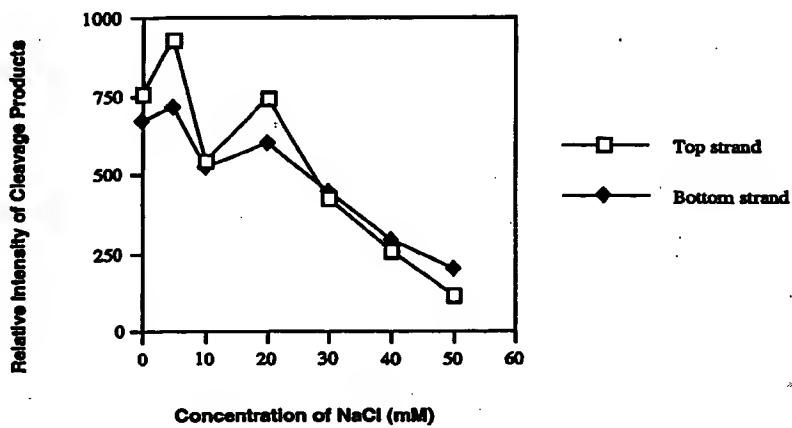


Figure 10

k-ras G12V (G->T)

A



B

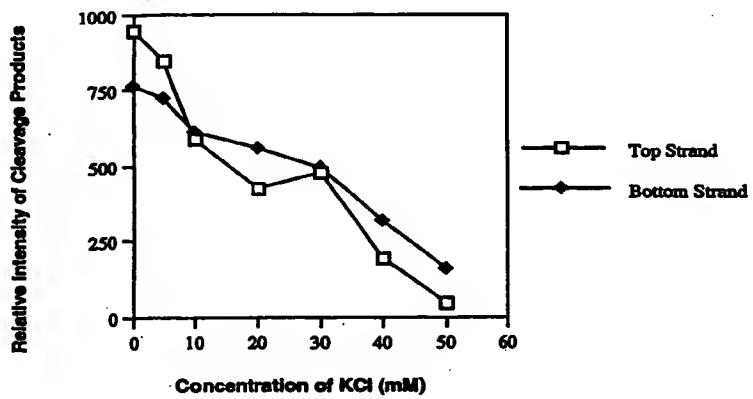


Figure 11

k-ras G12D (G->A)

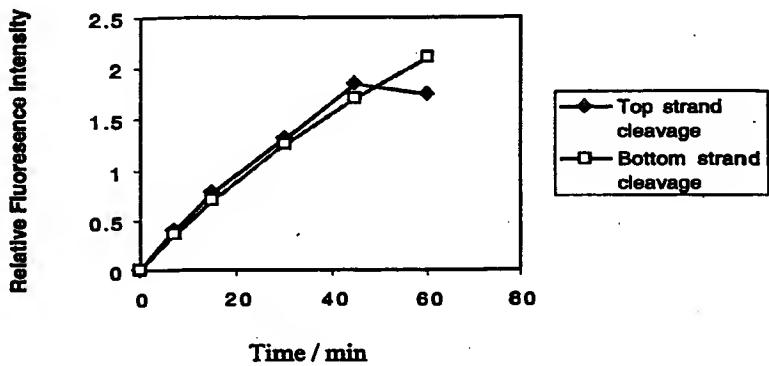
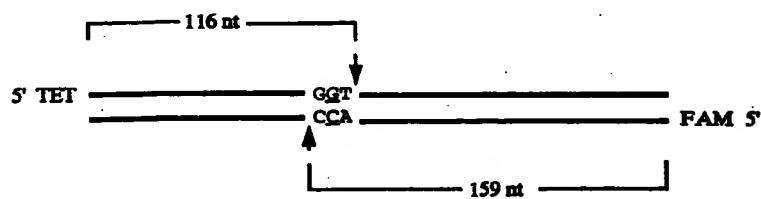


Figure 12

A



B

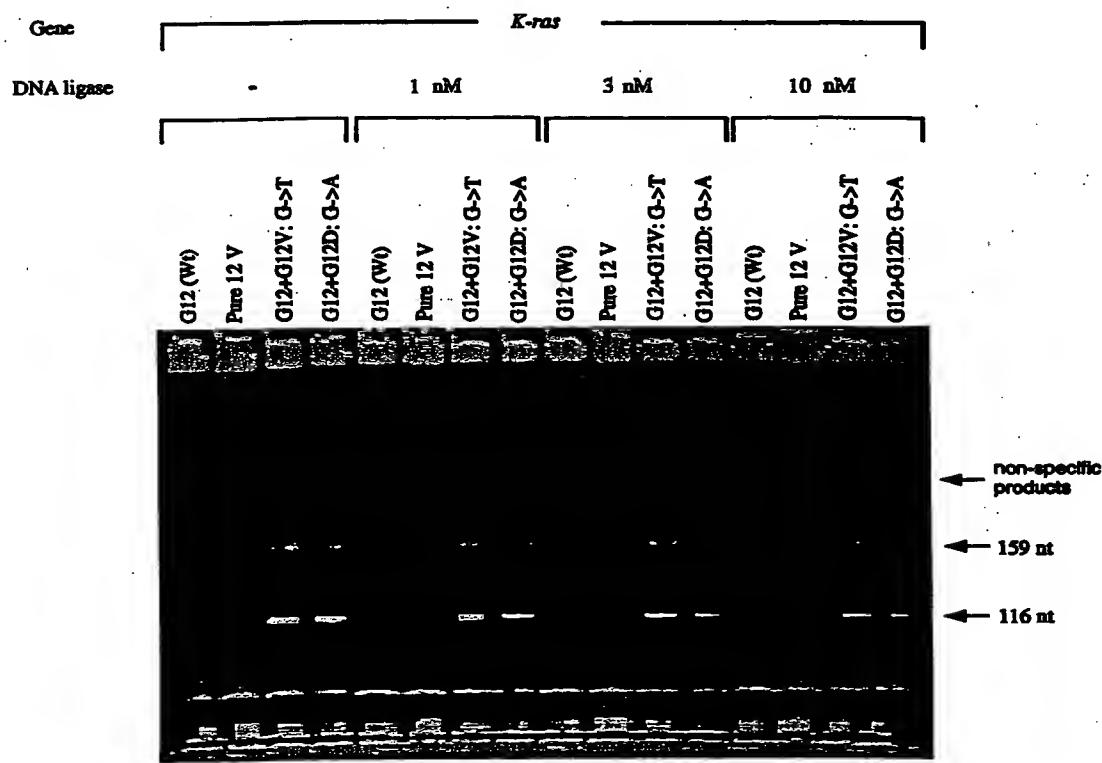


Figure 13

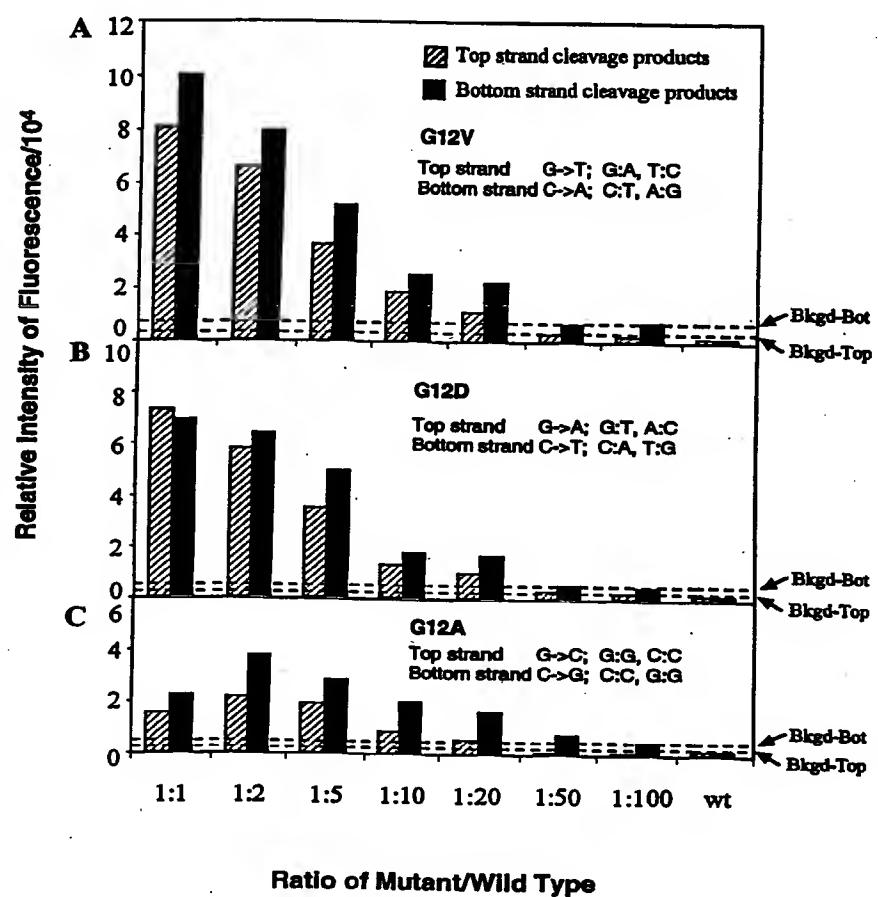


Figure 14

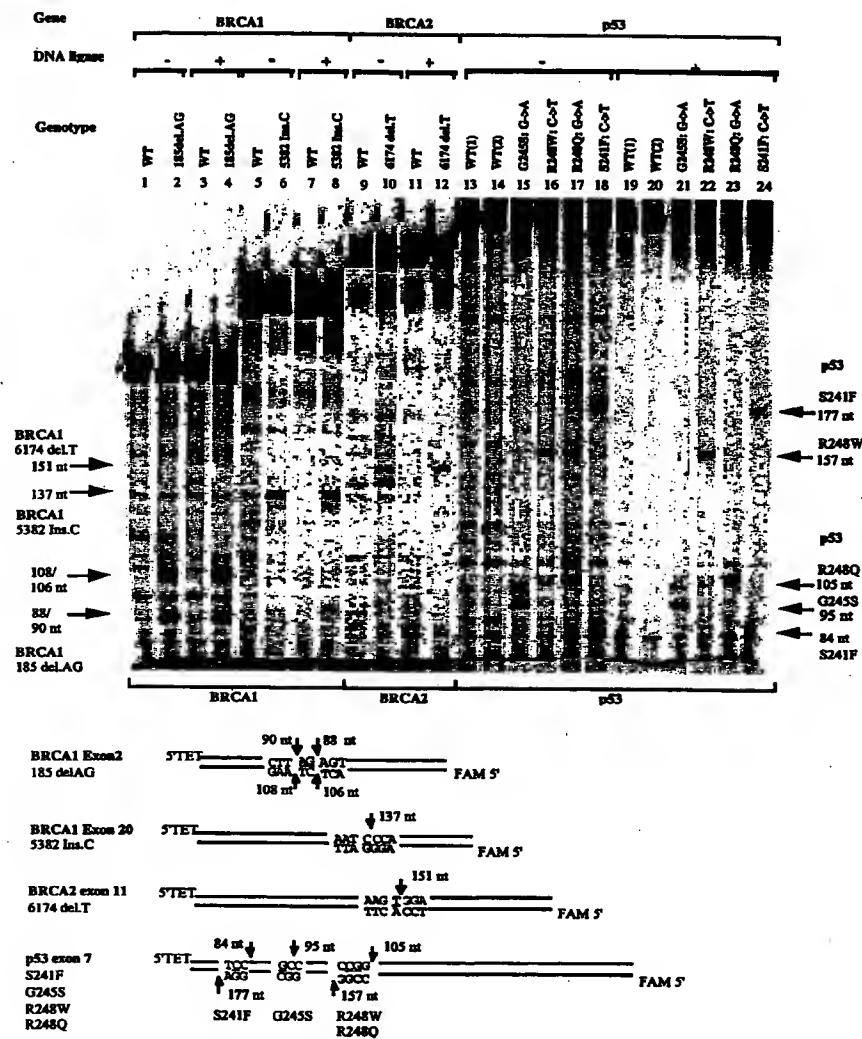


Figure 15

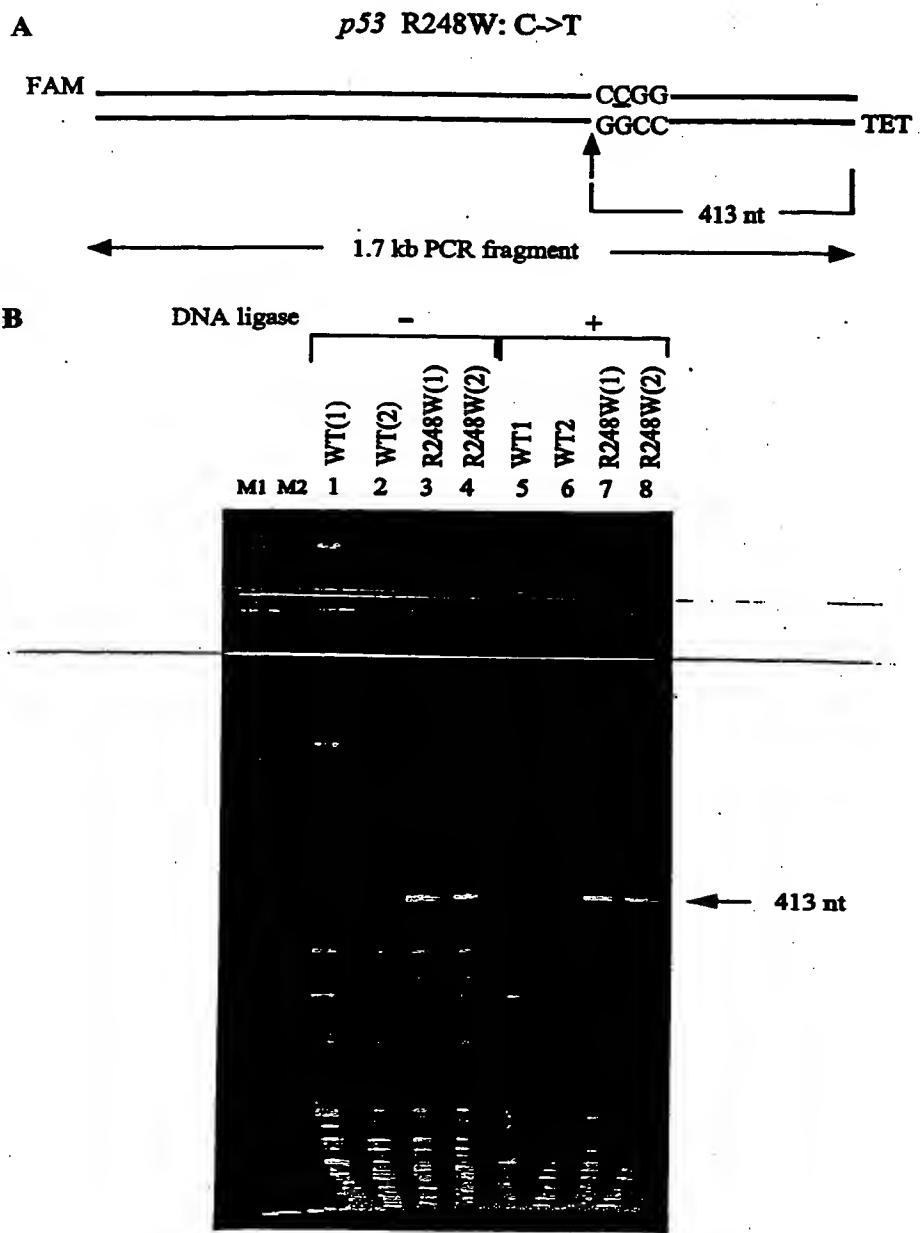
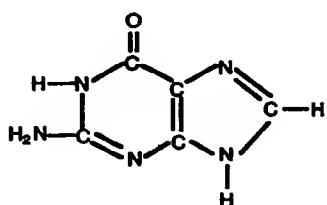


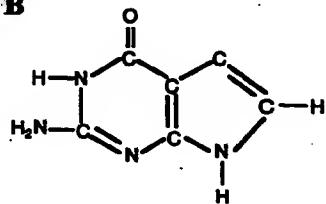
Figure 16

A



Guanine

B



7-deaza-Guanine

C

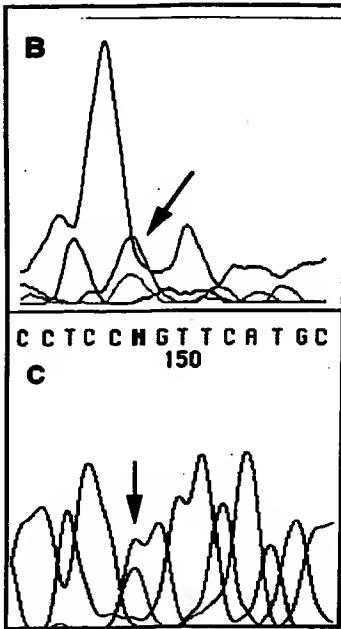
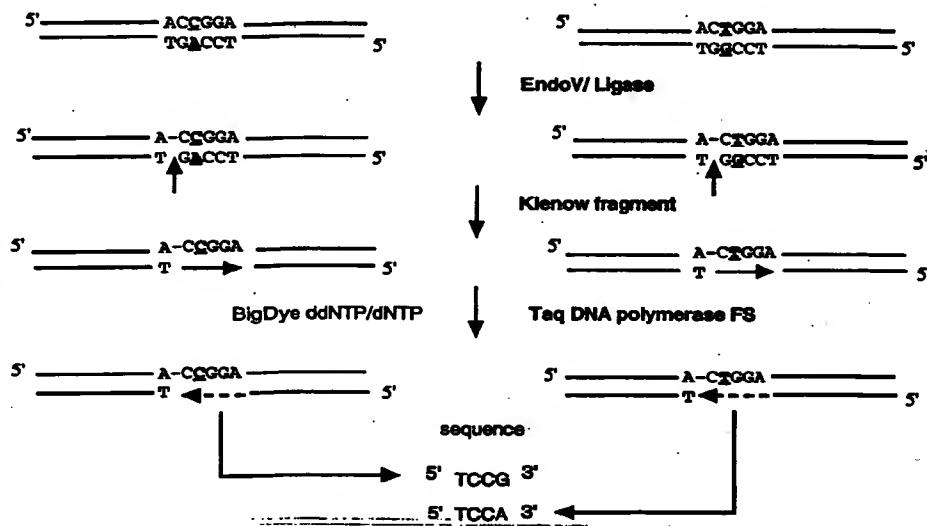
dG 7deazaG



Bottom strand
cleavage
products

Top strand
cleavage
products

Figure 17

A*p53 R248W(C>T)***Figure 18**

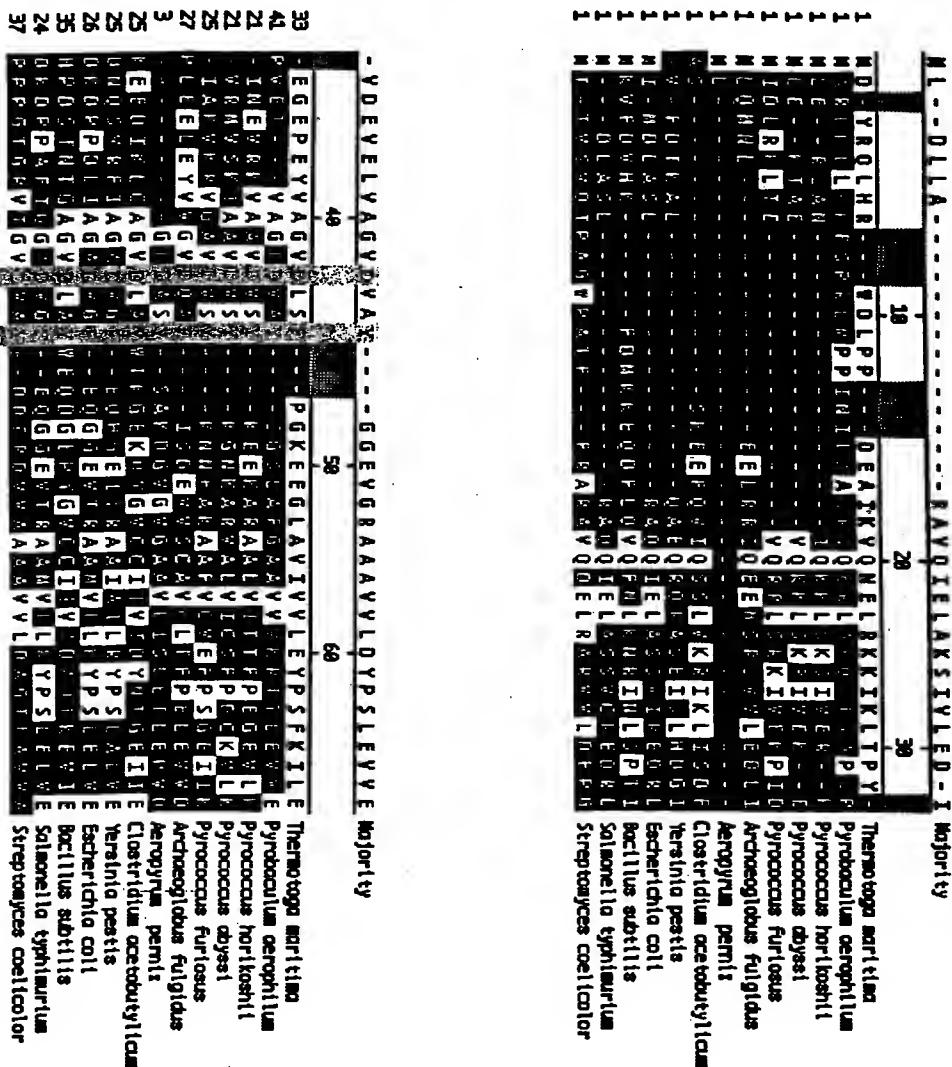


Figure 19

Figure 19 (cont.)

F--LED--GA--PLLOGGEGLGGYVLRK--REKPI--IVSYG Majority

	158	160	170
146	FRNPEOKBCSSSYLYDGEETTIGCVIRTKKEGSAP	I	VSPG
151	ED	D	Thermatopsporaristicia
125	LL	LL	Pyrobaculum aerophilum
125	LL	LL	Pyrococcus horikoshii
129	LL	LL	Pyrococcus abyssi
149	MWED	GLVLLDODGEIGIAGLAKRKEP	KAVSYSPG
189	TEPQON	Y	Pyrococcus furiosus
145	FENPE	FEDEAFLHIVNEEWIG	Pyrococcus furiosus
149	FELHQHODGAAQPLD	DHEEIGYVYK	Clostridium acetobutylicum
141	FEPF	LMDQHAYKAKRPEL	Vibrio cholerae
155	FHIEPE	ITIGEWEYG	Escherichia coli
139	FEPF	ALSPINODGEOL	Bacillus subtilis
152	DDPDT	PPRSITSPLAGAE	Salmonella typhimurium
		EWG	Streptomyces coelicolor

Block V

HRIILDSALAIVVAL-LD--GYBLPEPTRLADALAK-B-- Majority

	198	200	202	204	206	208	210	212	214	216	218	220	222
185	HLMOVES	S	K	LIA	KFTLP	-GRIP	EPTRLAH	-IYT	YRKL				
181	A	T	U	D	A	G	D	O	N	P	R	L	H
147	N	L	I	Y	I	E	A	I	R	A	I		
147	N	M	I	T	E	D	A	T	I	D	E		
151	M	L	I	L	D	A	R	I	W	E	L		
176	M	I	P	S	A	S	A	L	E	T	L		
142	H	R	I	T	E	F	A	R	I	P	-G		
184	H	R	I	D	E	F	A	R	I	P	-G		
179	H	R	Y	Y	S	A	A	W	U	S	-L		
188	H	R	Y	Y	S	A	A	W	U	S	-L		
194	H	R	Y	Y	S	A	A	W	U	S	-L		
178	H	R	Y	Y	S	A	A	W	U	S	-L		
191	H	R	Y	Y	S	A	A	W	U	S	-L		

Block VI

Figure 19 (cont.)

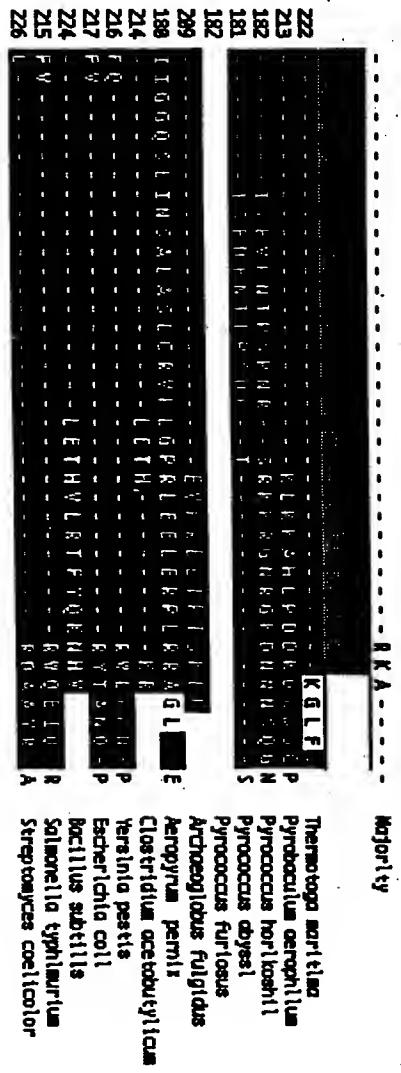


Figure 19 (cont.)

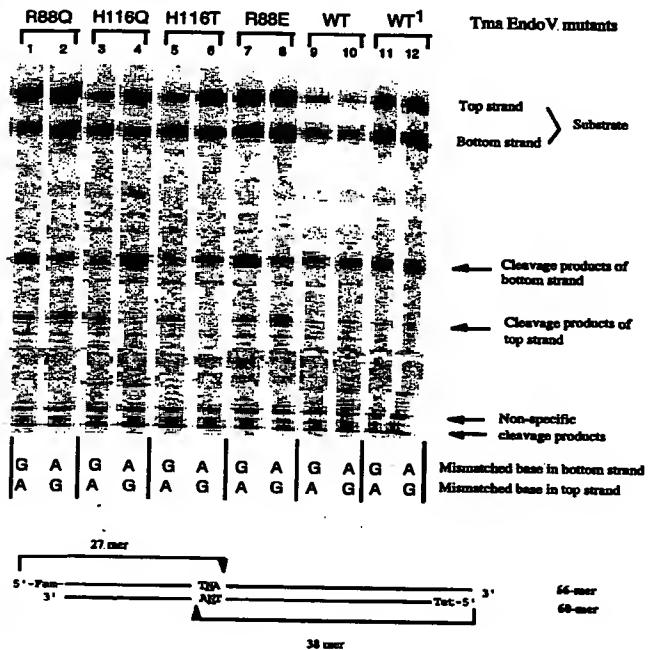


Figure 20

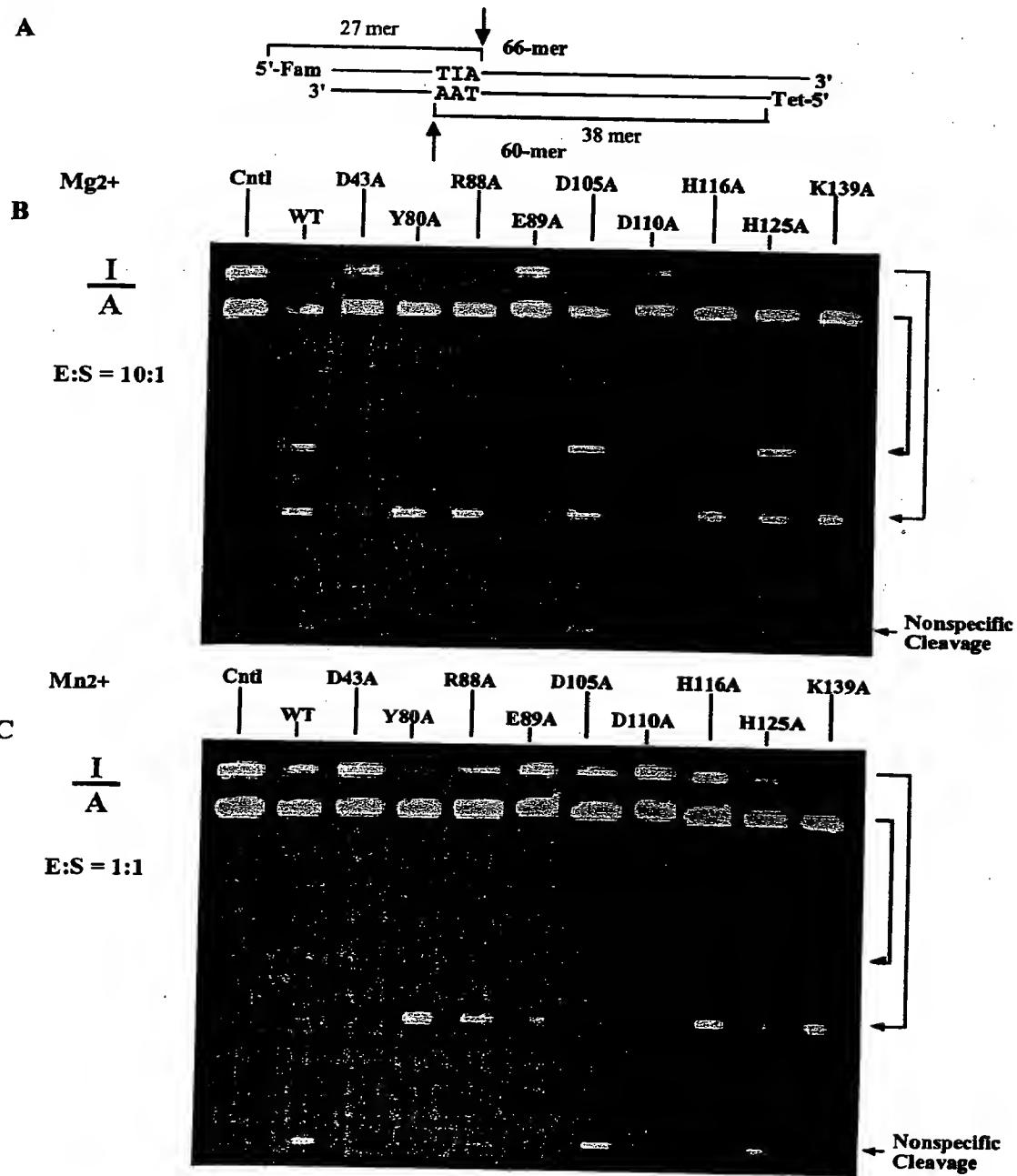


Figure 21

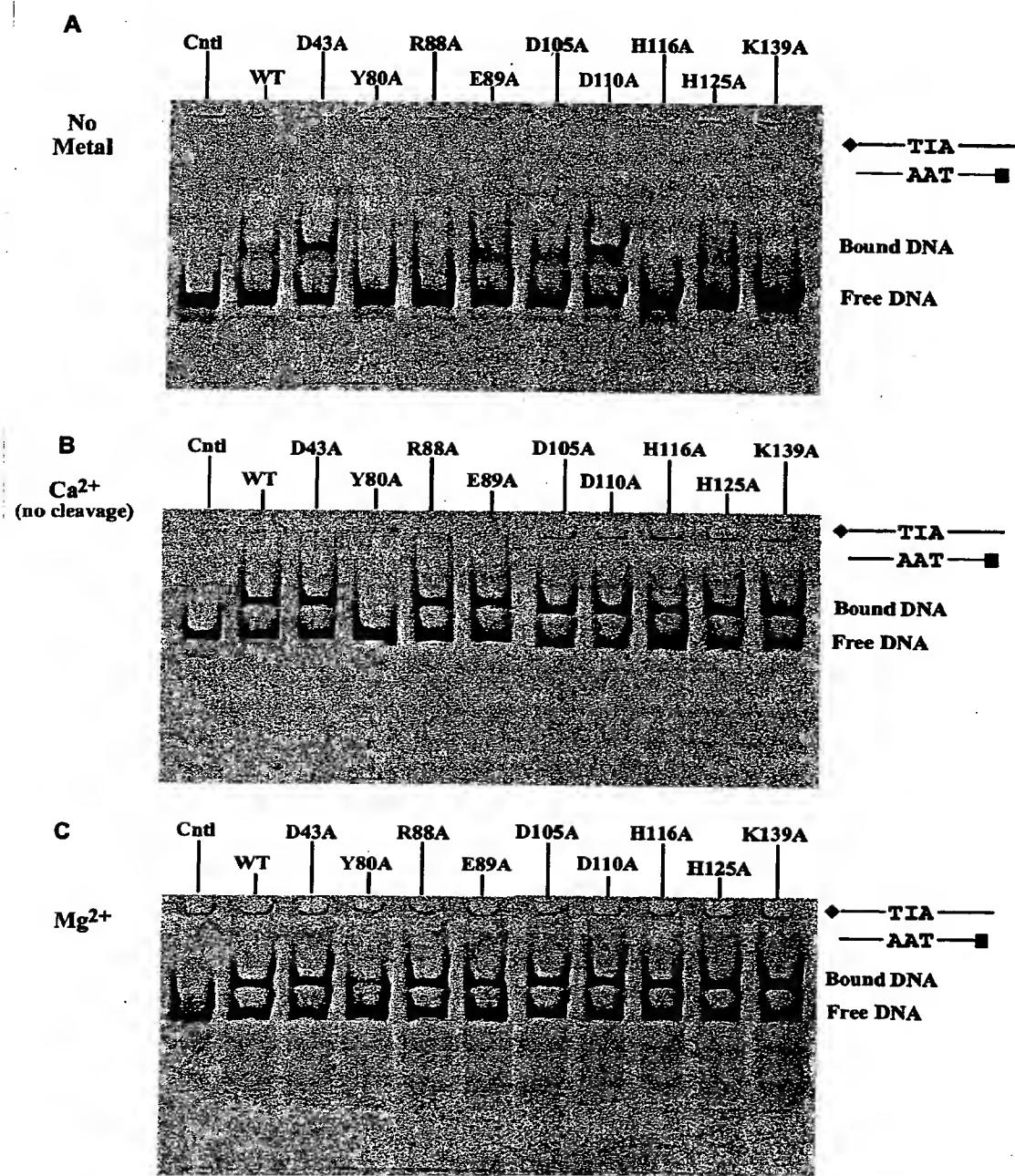


Figure 22

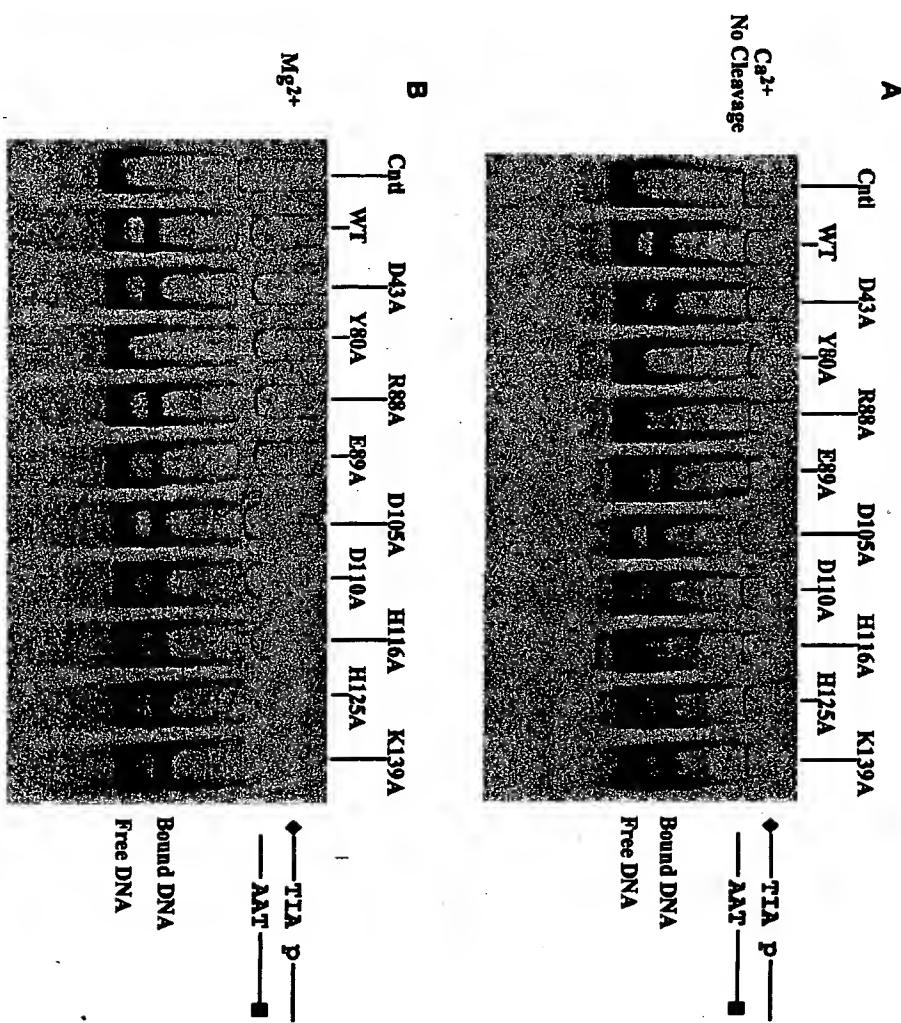
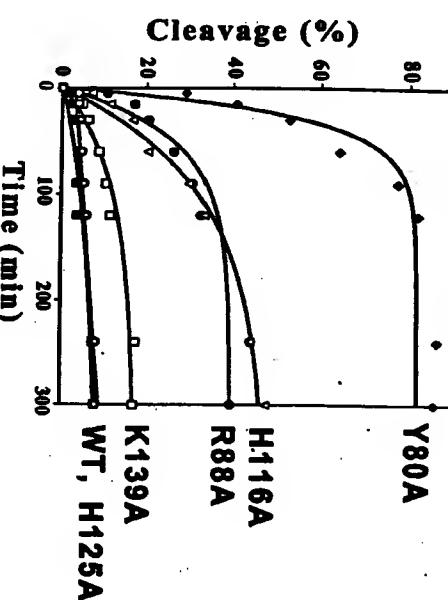


Figure 23

A Mg^{2+} E:S = 1:10

B
 WT
 $\text{—TIA—} + \text{O} \xrightarrow{\frac{k_1}{k_{-1}}} \text{—TIA—}_{\text{AAT}} \xrightarrow{k_2} \text{—TIA P—}_{\text{AAT}} \xrightarrow{k_3} \text{—TIA P—}_{\text{AAT}} + \text{O}$

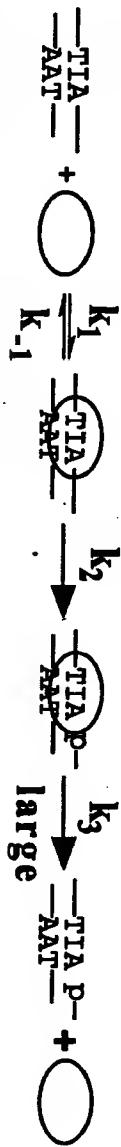
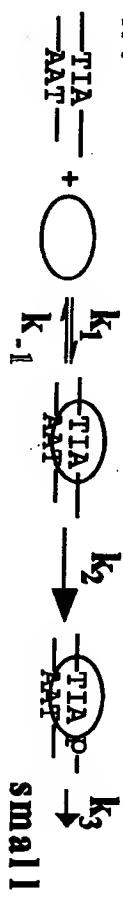


Figure 24

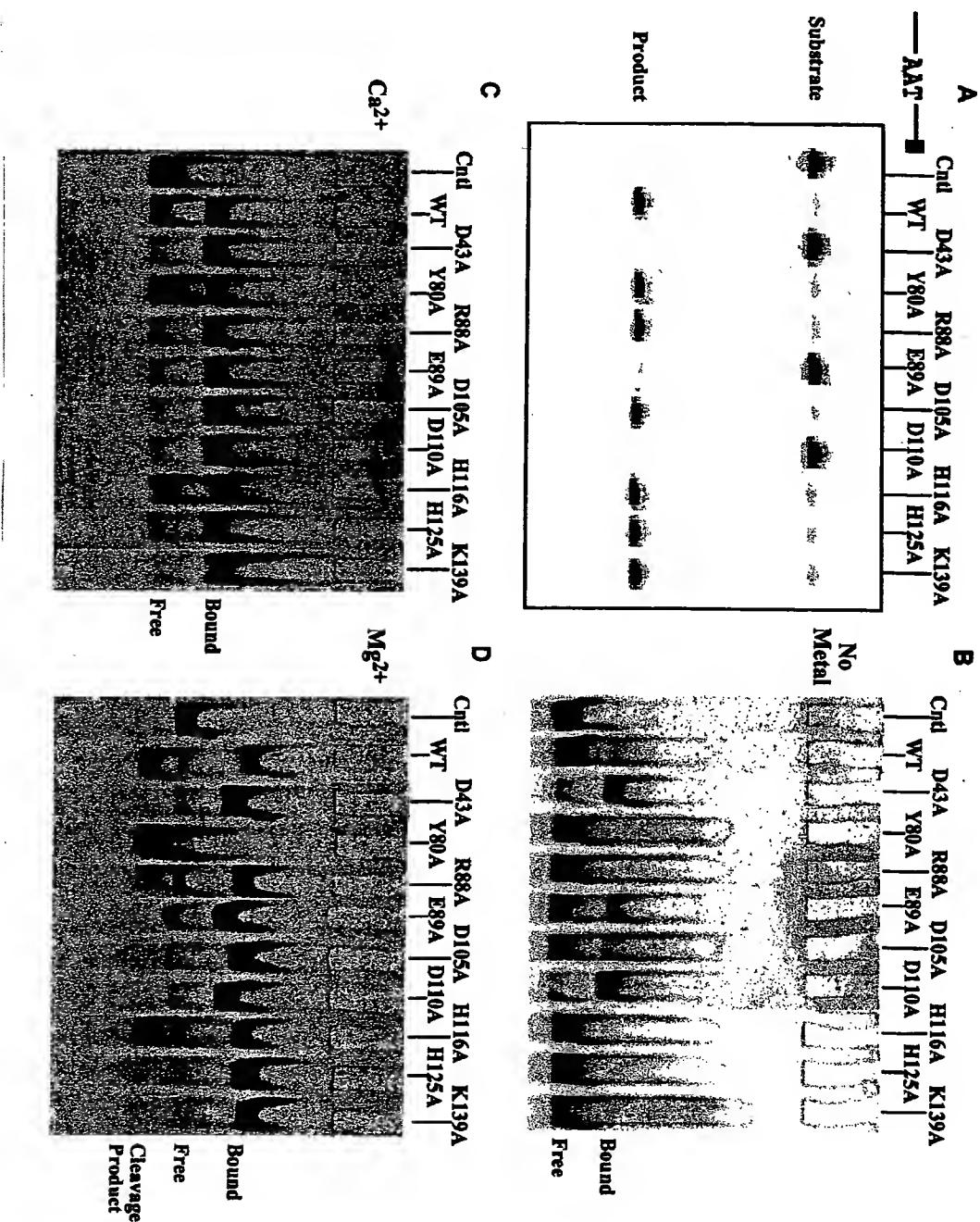


Figure 25

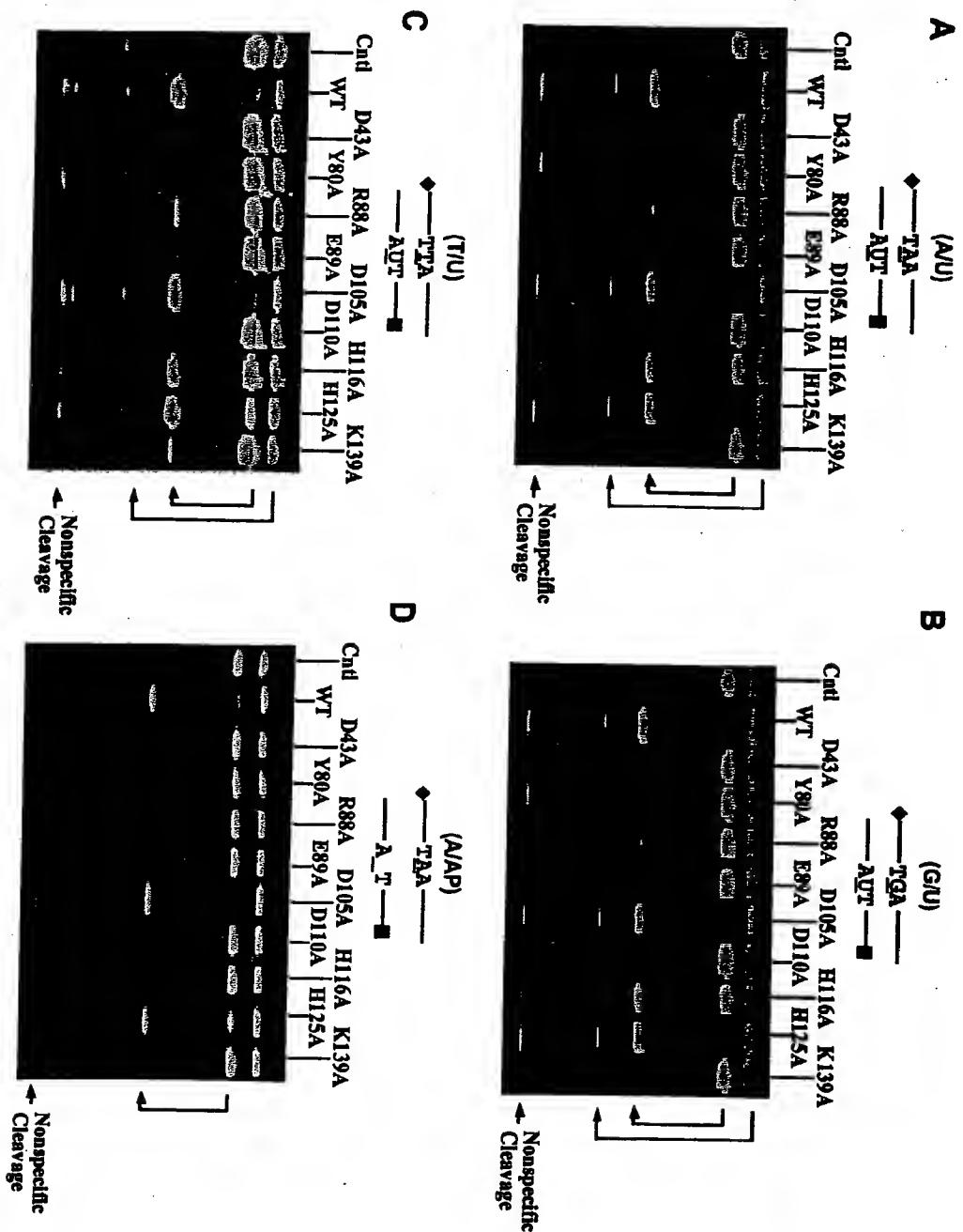


Figure 26